

## Changes in intervertebral disc morphology persist 5 mo after 21-day bed rest

Daniel L. Belavý,<sup>1</sup> P. Martin Bansmann,<sup>2</sup> Gisela Böhme,<sup>3</sup> Petra Frings-Meuthen,<sup>3</sup> Martina Heer,<sup>3</sup> Jörn Rittweger,<sup>3,5</sup> Jochen Zange,<sup>3,4</sup> and Dieter Felsenberg<sup>1</sup>

<sup>1</sup>Charité Universitätsmedizin Berlin, Centre of Muscle and Bone Research, Berlin; <sup>2</sup>KardioMR Köln-Bonn, Krankenhaus Porz am Rhein; <sup>3</sup>Institute of Aerospace Medicine, Deutsches Zentrum für Luft-und Raumfahrt; and <sup>4</sup>Medical Faculty of the University of Cologne, Cologne, Germany; and <sup>5</sup>Institute for Biomedical Research into Human Movement and Health, Manchester Metropolitan University, Manchester, United Kingdom

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**Belavý DL, Bansmann PM, Böhme G, Frings-Meuthen P, Heer M, Rittweger J, Zange J, Felsenberg D.** Changes in intervertebral disc morphology persist 5 mo after 21-day bed rest. *J Appl Physiol* 111: 1304–1314, 2011. First published July 28, 2011; doi:10.1152/jappphysiol.00695.2011.—As part of the nutrition-countermeasures (NUC) study in Cologne, Germany in 2010, seven healthy male subjects underwent 21 days of head-down tilt bed rest and returned 153 days later to undergo a second bout of 21-day bed rest. As part of this model, we aimed to examine the recovery of the lumbar intervertebral discs and muscle cross-sectional area (CSA) after bed rest using magnetic resonance imaging and conduct a pilot study on the effects of bed rest in lumbar muscle activation, as measured by signal intensity changes in T<sub>2</sub>-weighted images after a standardized isometric spinal extension loading task. The changes in intervertebral disc volume, anterior and posterior disc height, and intervertebral length seen after bed rest did not return to prebed-rest values 153 days later. While recovery of muscle CSA occurred after bed rest, increases ( $P \leq 0.016$ ) in multifidus, psoas, and quadratus lumborum muscle CSA were seen 153 days after bed rest. A trend was seen for greater activation of the erector spinae and multifidus muscles in the standardized loading task after bed rest. Greater reductions of multifidus and psoas CSA muscle and greater increases in multifidus signal intensity with loading were associated with incidence of low back pain in the first 28 days after bed rest ( $P \leq 0.044$ ). The current study contributes to our understanding of the recovery of the lumbar spine after 21-day bed rest, and the main finding was that a decrease in spinal extensor muscle CSA recovers within 5 mo after bed rest but that changes in the intervertebral discs persist.

magnetic resonance imaging; microgravity; spaceflight; low back pain; atrophy

WHILE OUR UNDERSTANDING OF the effect of prolonged bed rest (spaceflight simulation) on the lumbar intervertebral discs and musculature (3, 4, 6, 14, 21, 31) has improved greatly in recent years, our understanding of the recovery of these changes is less well developed. Commonly, studies consider the bed-rest phase alone, such as in the assessment of exercise countermeasures, with the subsequent recovery phase either not being examined or the data remaining unpublished. It is an unstated assumption that the various tissues of the human body recover after bed rest without specific intervention, although for a number of body systems, this assumption has not been tested.

Understanding these issues is important as, for example, prior work (3, 21) has linked the incidence of low back pain after bed rest to the extent of intervertebral disc changes and

muscle atrophy during bed rest. Similarly, in clinical studies of low back pain, alterations in disc dimensions have been linked to the recurrence of disc herniation (25) and muscle atrophy/dysfunction has been linked to the subsequent incidence of low back pain (18, 35). Such information is not only important ethically for bed-rest studies but is also relevant for understanding the increased incidence of lumbar intervertebral disc herniation seen in astronauts after spaceflight (23) and could also have applications in clinical situations.

In terms of the intervertebral discs of the lumbar spine, it is not clear when recovery occurs after bed rest. Seven weeks after a 17-wk bed rest, but not after a 5-wk bed rest, sagittal plane disc cross-sectional area (CSA) was shown to still be increased above baseline levels (28). Ninety days after a 60-day bed rest, disc volume, disc height, and spinal length remained greater than before bed rest (19). In another study, disc volume and anterior disc height remained greater than before bed rest in subjects scanned 90 days after the end of 90 days of bed rest (6). In yet a further study where disc morphology was measured up to 6 mo after 56 days of bed rest (4), it was unclear whether recovery of the discs occurred as baseline measurements were conducted on the first day of bed rest, after subjects had spent ~16 h in bed thus permitting increased changes in disc and spine morphology before baseline data was collected. Thus it is unclear to what extent recovery of the lumbar intervertebral disc occurs after bed rest.

In terms of the musculature of the lumbar spine, it is possible that the muscles recover their size spontaneously. In the musculature of the spine (4, 6) and lower quadrant (34, 43), partial recovery of muscle atrophy appears to occur 14 days after prolonged bed rest, with the process being complete by 90 days after bed rest. Seven weeks after a 17-wk bed rest (29), muscle volume in the spine and lower limb appeared to have recovered, but in this study data were available from two subjects only. Consequently, the aim of the current work was to gain a deeper understanding of the recovery of the lumbar intervertebral discs and lumbar musculature after bed rest as well as to track any relation these changes may have to low back pain incidence.

In addition to assessing morphological parameters of the lumbar intervertebral discs and muscles, the activation of the muscles by the central nervous system plays an important role in the stabilization of the lumbar spine (37). Magnetic resonance imaging (MRI) has been used previously to measure signal intensity changes in T<sub>2</sub>-weighted MRIs of the musculature after exercise as a measure of muscle function and activation levels of the calf (26) and thigh (16, 42) musculature. Thus an additional goal was to perform a pilot study examining the effects of bed rest on muscle activation, as measured by

Address for reprint requests and other correspondence: D. L. Belavý, Zentrum für Muskel-und Knochenforschung, Charité Campus Benjamin Franklin, Hindenburgdamm 30, D-12200 Berlin, Germany (e-mail: belavy@gmail.com).

signal intensity changes in T<sub>2</sub>-weighted magnetic resonance images, in the lumbar spine musculature after a spinal extension exercise.

The “nutrition countermeasures” (NUC) 21-day bed rest study provided the opportunity to assess these issues. The primary goal of this study was to examine the effect of a nutrition countermeasure, which involved potassium bicarbonate, on preventing increased bone resorption during bed rest. Effects on muscle CSA, muscle function, and the intervertebral discs of this countermeasure were not expected, however, and the study presented an opportunity to better assess the effects of bed rest and recovery on the lumbar spine.

## MATERIALS AND METHODS

*Bed-rest protocol, subject characteristics, and nutrition protocol.* Seven healthy male subjects (Table 1) were recruited for participation in the nutrition countermeasures (NUC) bed rest study at the German Aerospace Center in Cologne, Germany in 2010. Exclusion criteria relevant for the current study included professional athletes, claustrophobia, metal implants, muscle or joint disease, and prior history of intervertebral disc protrusion.

Two campaigns (C1 and C2) were conducted and all subjects participated in both campaigns as part of a crossover design. Each campaign consisted of 7 days of prebed-rest baseline data collection (BDC-7 to BDC-1), 21-days of strict 6° head-down tilt bed rest (HDT1 to HDT21), and 6 days of postbed-rest recovery (R + 0 to R + 5). Reambulation occurred on the morning of R + 0. Subjects also returned to the facility on R + 14 and R + 28 for follow-up assessment. The second bed-rest phase (C2 HDT1) began 154 days after the end of the first campaign (C1 R + 0). The study was approved by the ethical committee of the Aerztekkammer Nordrhein, Duesseldorf, Germany, and subjects gave their informed written consent.

As part of the crossover design, a nutrition countermeasure involving sodium bicarbonate was trialed, but as no significant effects on muscle size or the intervertebral discs were expected or observed, the results from both groups have been pooled. The primary outcome measure of the NUC bed-rest study was bone resorption markers from 24-h urine collections and not the parameters of the current study. Assuming a power of 0.8, an  $\alpha$ -level of 0.05, and given an SD of 3.7% for within-subject differences in change in average lumbar erector spinae muscle CSA and 4.5% for average lumbar intervertebral disc volume as well as a 0.96 (erector spinae CSA) and 0.98 (disc volume) correlation between repeated measures (based on prior data after 28-day bed rest from our group; Ref. 2), an effect size between groups of 2.5% for erector spinae CSA and 2.5% for intervertebral

disc volume should be detected given seven subjects in each group in this crossover design with three repeated measures. G\*Power (version 3.1.2; <http://www.psych.uni-duesseldorf.de/abteilungen/aap/gpower3/>) was used for these calculations.

*Physical activity and low back pain questionnaires.* In the BDC phase of each campaign and on R + 28 of the second campaign, subjects completed a questionnaire on habitual physical activity (1). This physical activity questionnaire has been validated previously (1) and assesses occupational (e.g., time spent standing, sitting, lifting heavy items an so forth at work), sport related (i.e., types of sports played, how frequently, how long), and leisure time (e.g., extent of television viewing) physical activity. For the analyses presented here, we focused on total physical activity, which represents the summation of the “work,” “sport,” and “leisure” indexes.

Subjects also completed a pain questionnaire twice before bed rest (BDC-5 and BDC-2), on the first 7 days (HDT1 to HDT7) as well as days 10, 13, 16, 19, and 21 (HDT10, HDT13, HDT16, HDT19, and HDT21) of bed rest, and on every postbed-rest recovery day (R + 0 to R + 6, R + 14, and R + 28). Subjects indicated whether they had any pain or discomfort in any body region, and if so, this region was marked as well as its intensity on a 100-mm visual analog scale (Ref. 15). Incidence of low back pain was defined as any report of pain or discomfort between the first lumbar vertebrae and the coccyx. Before the beginning of the bed rest phase in C1, subjects were also questioned regarding prior history of musculoskeletal pain and injury.

*MRI and loading protocol.* MRI was performed during the baseline data collection (BDC) phase, 2 days before bed rest (BDC-2; beginning at 2 PM), and then on the first day of postbed-rest recovery (R + 1; beginning at 10 AM) and then again on the fifth day of postbed-rest recovery (R + 5; beginning at 2 PM). The same schedule was used in both C1 and C2, and 147 days elapsed between C1 R + 5 and C2 BDC scanning, implying that C2 BDC was equivalent to C1 R + 153. Five days before bed rest in each campaign, subjects were familiarized with the loading protocol.

A 1.5T Siemens Sonata (Erlangen, Germany) MR scanner at the Krankenhaus Porz am Rhein, an external hospital located nearby, was used for all scanning sessions. At every measurement sitting, after subjects spent 1 h 15 min in supine lying while MR scanning was performed as part of other experiments, the following protocol was used:

1) The subject was positioned in supine lying with a standard support behind his knees. After initial pilot scanning, 29 sagittal images (thickness 3 mm; interslice distance: 0.3 mm; repetition time: 6,220 ms, echo time: 105 ms, field of view: 340 × 340 mm interpolated to 384 × 384 pixels) were taken to encompass the entire vertebral body and include the transverse processes from the lower thoracic spine (typically T<sub>10</sub>) to the sacrum (Fig. 1).

Table 1. *Subject characteristics*

Subject	Weight, kg	Age, yr	Height, cm	Group in C1	Total Physical Activity		
					C1 BDC	C2 BDC	C2 R + 28
A	85.2	27	185	PoBi	10	9.25	8.875
B	70.1	25	182	Ctrl	9.75	9.25	8.375
C	85.1	29	178	PoBi	7.875	9.125	8.75
D	73.8	22	179	Ctrl	6.75	8.375	7.25
F	84.8	32	186	Ctrl	8.375	8.875	8.375
G	76.4	26	179	PoBi	7.625	7.75	7.25
H	72.3	30	176	Ctrl	7.375	8	9
Mean(SD)	78.9 (6.4)	30 (7)	181 (4)		8.2 (1.1)	8.7 (0.6)	8.3 (0.7)

In the 2nd campaign (C2) each subject participated as his own control in the other group as part of the crossover design. Data on age, height, and weight are from subjects in the first campaign (C1) 7 days before the beginning of bed rest. Total physical activity score refers to data from Baecke habitual physical activity questionnaire (Ref. 1; no units) completed before bed rest (BDC) in both the 1st (C1) and 2nd (C2) campaign as well as 28 days after the end of the bed rest phase of C2. Total physical activity did not differ significantly between measurement days ( $P = 0.38$ ). PoBi, standard nutrition plus potassium bicarbonate group; Ctrl, standard nutrition group. See RESULTS for further details.



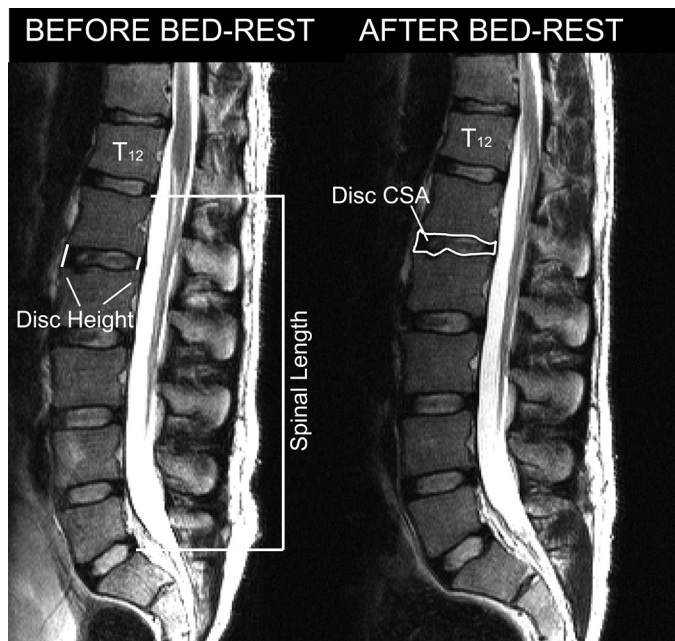


Fig. 1. Measurements of disc and spine morphology. *Left*: before bed rest; *right*: first day after bed rest in same subject. Anterior and posterior disc height was of each vertebral disc from T<sub>12</sub>L<sub>1</sub> to L<sub>5</sub>S<sub>1</sub> (shown at L<sub>2</sub>L<sub>3</sub> at *left*). Disc volume was interpolated from sagittal plane disc cross-sectional area (CSA) measurements (shown at L<sub>2</sub>L<sub>3</sub> at *right*). Spinal length, parallel to scanning table, was measured between the dorsorostral corner of S<sub>1</sub> and the dorsorostral corners T<sub>12</sub>, L<sub>1</sub>, L<sub>2</sub>, L<sub>3</sub>, L<sub>4</sub>, and L<sub>5</sub>. Intervertebral length was then calculated between each vertebra. While images positioned at the spinous process are presented here, all available images were measured and the average of disc heights and intervertebral length at each vertebral level and time-point was taken for each subject before further analysis. Increase in disc size after bed rest is particularly noticeable in this subject with a 30.7% increase in the volume of the L<sub>5</sub>S<sub>1</sub> intervertebral disc.

2) Then, five groups of three T<sub>2</sub>-weighted images each (slice thickness: 4 mm, interslice distance: 0 mm, repetition time: 3,500 ms, echo time: 107 ms, field of view: 260 × 235.62 mm interpolated to 384 × 348 pixels) were positioned over the center of each vertebral body from L<sub>1</sub> to L<sub>5</sub> and to then angulated to be parallel to the superior vertebral endplate of each vertebra (Fig. 2).

3) The subject was taken out of the MR scanner, remained in lying, and was transferred to a custom-built table that permitted free movement of the upper body while allowing bracing of the hips and legs. The subject was positioned in side lying, and a vest was brought about the subject's chest and shoulders to permit the application of a horizontally directed force to the subject's trunk. With external loading applied horizontally, the subject would then be required to generate a trunk extension force. A digital weight gauge (Votcraft HS-100; Conrad Electronic SE, Hirschau, Germany) was attached to the vest around the subject's chest via carabineers. Load was then applied with an initial ramp period of 6 s and the static holding period of 7 s. Ten repetitions were performed with a 20-s pause in between repetitions. In the first campaign (C1), load was set to 10% of subject body weight, and in the second campaign (C2), load was set to 30% of subject body weight. The average body weight from the BDC phase of the first campaign (Table 1) was used for loading level calculation. Ideally, both loading levels should have been performed during the same testing session in both campaigns, but due to time restrictions, this was not possible. As it was preferred to avoid high levels of spinal loading immediately after bed rest, these loading levels were chosen based on pilot trials that showed increases in spinal extensor signal intensity with these lower loads. The timing of loading and testing duration were monitored with custom written software in the Labview

6 environment (National Instruments). The same operator (D. L. Belavý) conducted all loading sessions.

4) At the end of the loading protocol, the subject was returned immediately to the MR scanner and the sagittal sequence from *step 1* was performed again followed by the para-axial sequence from *step 2*. The mean(SD) duration between the end of the exercise and the beginning of sagittal scanning was 4.0(0.7) min and another 4.7(0.4) min elapsed before the para-axial sequences began.

At the end of each scanning session, data were then stored for offline processing.

**Blinded image measurements.** To ensure measurer blinding, each data set was assigned a random number (www.random.org). ImageJ 1.39u (<http://rsb.info.nih.gov/ij/>) was used for MRI analyses that were conducted by the same operator (D. L. Belavý). The following measures of spinal morphology were conducted, as in prior work (2, 3), in every image where the required anatomical landmarks could be delineated (Fig. 1): 1) disc volume of each disc from T<sub>12</sub>L<sub>1</sub> to L<sub>5</sub>S<sub>1</sub> was interpolated from all sagittal plane CSA measures of each disc; 2) anterior and posterior disc height were measured from T<sub>12</sub>L<sub>1</sub> to L<sub>5</sub>S<sub>1</sub>; and 3) intervertebral length, the vertical distance between the dorsorostral corners T<sub>12</sub>L<sub>1</sub> to L<sub>5</sub>S<sub>1</sub>, was measured. Since multiple measurements (disc volume: median[minimum-maximum] of 13[9–17] measures per disc, subject and time point; disc height: 12[9–16]; intervertebral length: 12[8–16]) were made, the average value of all measurements for disc heights and intervertebral length at each vertebral level and time-point for each subject was taken before further analysis. In contrast to prior work (2, 6), data on intervertebral angles and lumbar lordosis were not included, as no significant effects of bed rest, recovery, or spinal loading were seen.

Bilateral CSA measurements of the lumbar multifidus, erector spinae, quadratus lumborum, and psoas muscles were conducted on the para-axial MRI (Fig. 2). To accurately delineate the multifidus muscle and the more laterally placed longissimus muscle, the fascial border (11) separating these two muscles was used as an anatomical landmark. Signal intensity was also measured in the same regions of interest as per muscle CSA measurements.

**Further data processing and statistical analyses.** Muscle CSA data were averaged between each of the three images at each vertebral level on each side and then averaged between left and right sides. For both muscle CSA data and disc and spine morphology as no significant effects were found on these parameters due to loading, the

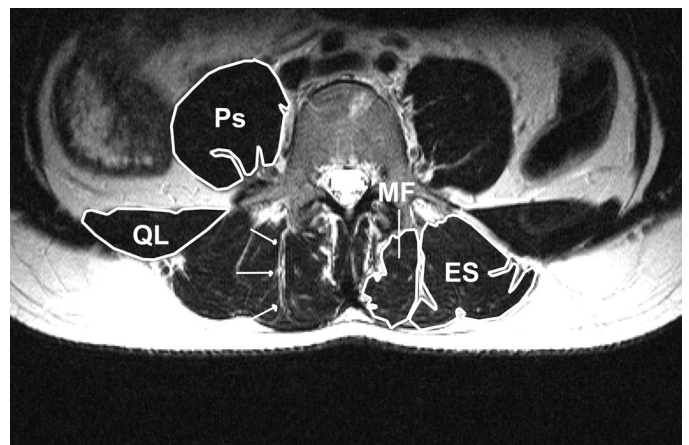


Fig. 2. Muscle CSA measurements. CSA measurements were made of the psoas (Ps), erector spinae (ES), and multifidus (MF) muscles from L<sub>1</sub> to L<sub>5</sub>. Quadratus lumborum (QL) was measured from L<sub>1</sub> to L<sub>4</sub> as it was typically absent at L<sub>5</sub>. Arrows indicate the fascial border between MF and ES that aided delineation of these 2 muscles. Signal intensity was also measured in the same regions of interest. Three images were obtained from each vertebral level oriented parallel to the superior vertebral endplate. Here is an example para-axial image from L<sub>4</sub>.

measurements from before and after loading were averaged to reduce measurement error. For each of these variables, linear mixed-effects models (40) were used to model main effects of study date and intervertebral level as well as their interaction. Random effects for each subject and, where necessary, intervertebral-level within subject was modeled and where necessary allowances for heterogeneity of variance (such as due to intervertebral-level or study-date) were permitted. ANOVA then evaluated the significance of each of the model parameters and, where appropriate, subsequent a priori contrasts compared first campaign BDC values to values from subsequent testing dates. To evaluate the relationship between changes in muscle CSA and disc and spine morphology, partial correlation analyses, controlling for study date, were also performed.

For evaluation of muscle signal intensity changes due to loading, signal intensity measurements were averaged, weighted by CSA, between images at the same level on the same side of the body and then between sides of the body. Signal intensity, averaged across all intervertebral levels weighted by CSA, was evaluated in statistical analyses, although data from each vertebral level were also considered. Similar linear mixed-effects models were used with appropriate main-effects, interactions, random effects, and allowances for heterogeneity of variance.

To evaluate the impact of the nutrition countermeasure and relationship to low back pain after bed rest ( $R + 0$  to  $R + 28$ ), the data were averaged across lumbar intervertebral-levels and the percentage change compared with before bed rest (BDC) calculated. For low back pain, the relationship to changes on  $R + 1$  only were considered. Linear mixed-effects models were similarly used for these analyses.

Differences between the bed-rest and recovery phases in the low back pain data were considered. The Mann-Whitney  $U$ -test was used for intensity of pain and duration of pain, and Fisher's exact test was used for the number of subjects reporting pain. An  $\alpha$  of 0.05 was taken for statistical significance and the "R" statistical environment (version 2.10.1, [www.r-project.org](http://www.r-project.org)) was used for all analyses. Unless otherwise stated, all data are reported as mean(SD).

## RESULTS

All subjects completed all testing dates as planned. As expected, no significant effects were seen of the nutrition countermeasure on the parameters of the current study ( $P \geq 0.22$ ). Data on total physical activity from questionnaires were similar ( $P = 0.38$ ; Table 1) among the BDC phase of C1, the BDC phase of C2, and 28 days after bed rest ( $R + 28$ ) in C2. Analyzing the physical activity data separated into the work, sport, and leisure indexes gave similar results ( $P \geq 0.13$ ; data not shown).

**Low back pain incidence.** One subject reported an 11-yr history of low level chronic low back pain subsequent to a lumbar spine fracture. None of the remaining subjects reported any prior history of low back pain. During bed rest, the incidence of low back was highest in the first 4 days and then reduced towards the end of bed rest. After the subjects reambulated, the incidence of low back pain increased (Fig. 3). The data on pain intensity are presented in Fig. 3. All reports of low back pain were located centrally at the lumbar spine, with no reports of unilateral pain, pain radiation into the extremities, parasthesia, or anesthesia.

In the HDT phase of the C1, six subjects reported pain of duration from 1 to 7 days. In the subsequent recovery phase, three subjects reported 1 day of pain and one subject reported 5 days of pain. In the HDT phase of the second campaign, four subjects reported pain on 1 to 4 days, but one subject reported pain on 11 of 12 measurement dates. In the recovery phase of the second campaign, three subjects reported low back pain

from 2 to 4 of the 8 measurement days. The intensity of pain, duration of pain, and number of subjects reporting pain were statistically similar ( $P$  all  $\geq 0.43$ ) between HDT and recovery phases as well as between campaigns.

**Intervertebral disc and spine morphology.** Baseline, first campaign BDC, data are given in Table 2. ANOVA of the data from all intervertebral levels showed a significant study-date main effect for anterior disc height ( $P = 0.006$ ), posterior disc height ( $P = 0.030$ ), disc volume ( $P < 0.001$ ), and intervertebral length ( $P < 0.001$ ), with significant differences between vertebral levels in their response over time for anterior disc height ( $P = 0.012$ ), posterior disc height ( $P = 0.021$ ), disc volume ( $P = 0.006$ ), and intervertebral length ( $P = 0.008$ ). Disc volume increased the most at the lower lumbar spine with progression down to decreases in disc volume at  $T_{12}L_1$  (Fig. 4). A similar pattern was seen for intervertebral length (Fig. 4) and disc heights (Fig. 5). Additional analyses showed no significant differences between  $R + 1$  and  $R + 5$ . Also, further analysis showed that between the end of the first campaign ( $R + 5$ ) and 147 days later at second campaign baseline, no significant changes were seen in any of the parameters with the exception of intervertebral length at  $L_4L_5$  ( $P = 0.014$ ). If first campaign  $R + 1$  is chosen for comparison with second campaign BDC, then only intervertebral length at  $L_2L_3$  shows some reductions ( $P = 0.042$ ). When the average of all lumbar values was considered, no significant differences between the end of the first campaign and second campaign BDC were seen. The extent of intervertebral disc and spine morphology changes on  $R + 1$  and  $R + 5$  compared with precampaign BDC did not differ between the two campaigns ( $P \geq 0.16$ ). No relationship was seen between the extent of disc and lumbar morphology changes on  $R + 1$  and the incidence of low back pain between  $R + 0$  and  $R + 28$  ( $P \geq 0.08$ ).

**Muscle CSA.** All muscles ( $P < .001$ ) except psoas ( $P = 0.10$ ) showed a significant study-date main effect on ANOVA, whereas psoas and multifidus showed a significant study-date  $\times$  intervertebral-level interaction on ANOVA ( $P < .001$ ; Table 3). Erector spinae, multifidus, and quadratus lumborum showed reductions in CSA after bed rest, which tended to be greater in the upper lumbar region in the erector spinae; however, significant increases in psoas CSA at the lower lumbar spine were seen after bed rest in the first campaign and also on  $R + 5$  after the second campaign. One-hundred and forty-seven days later at BDC scanning in the second campaign, muscle CSA had returned to prebed-rest levels in all muscles, although CSA was significantly greater than before the first campaign at the lower lumbar levels of multifidus, psoas, and quadratus lumborum (Table 3). Between  $R + 1$  and  $R + 5$  (pooled across both campaigns), significant increases in muscle CSA were seen in the erector spinae at  $L_2$ ,  $L_3$ , and  $L_4$  ( $P \leq 0.05$ ), multifidus at  $L_4$  and  $L_5$  ( $P < 0.001$ ), psoas at  $L_5$  ( $P = 0.021$ ), and quadratus lumborum at  $L_2$  and  $L_3$  ( $P = 0.048$ ). Similar to spinal morphology data, the percentage change of muscle CSA changes after bed rest on  $R + 1$  and  $R + 5$  compared with precampaign BDC did not differ between the first and second campaigns ( $P \geq 0.84$ ). Partial correlation analyses between muscle CSA changes and disc and spinal morphology changes are reported in Table 4. Subjects who reported low back pain between  $R + 0$  and  $R + 28$  showed greater losses of multifidus CSA ( $P = 0.044$ ) on  $R + 1$  than those who did not and showed reductions in psoas CSA rather than increases seen in the other

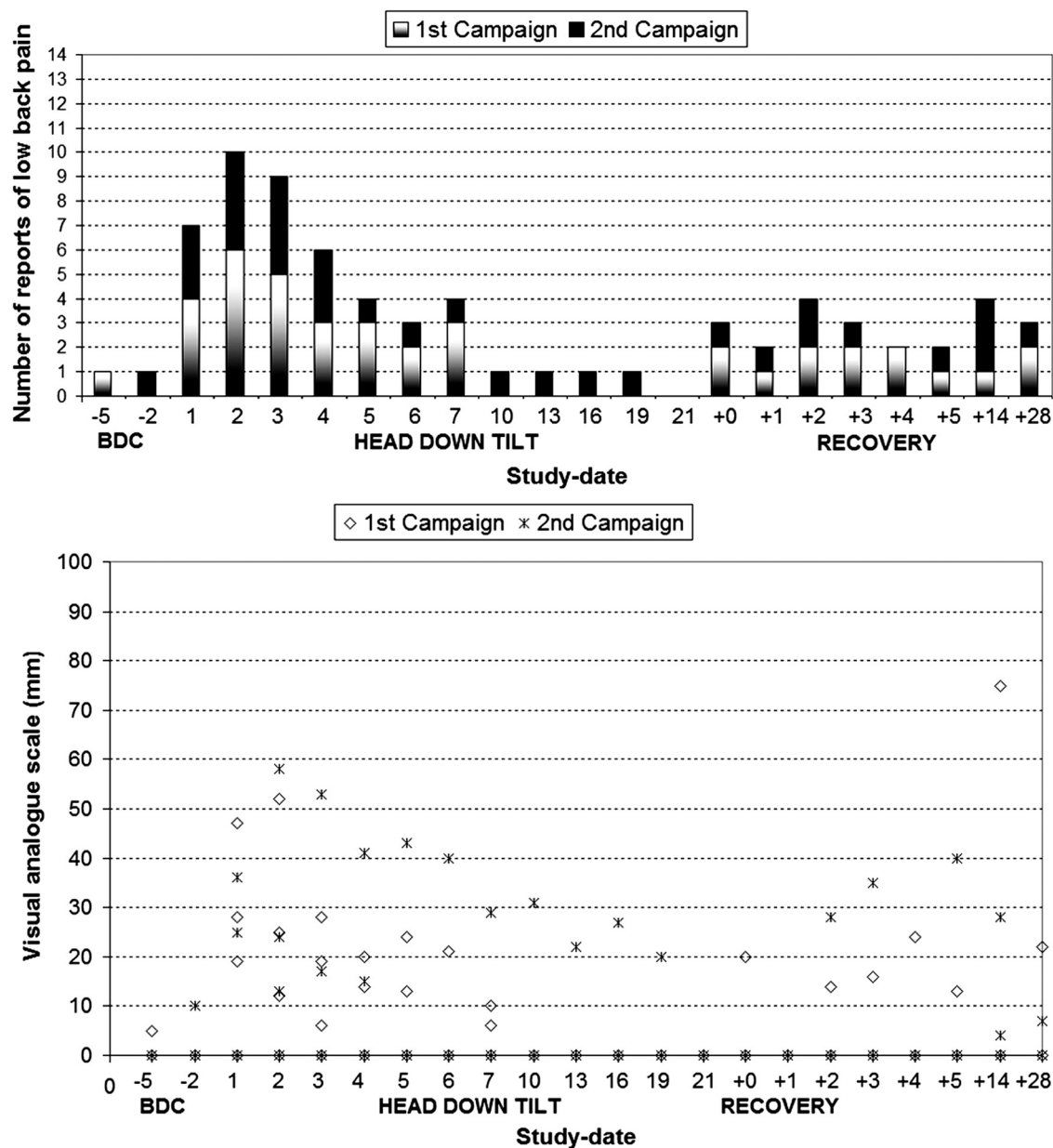


Fig. 3. Low back pain incidence (*top*) and severity (*bottom*) during and after bed rest. Values at *bottom* are visual analogue scale pain intensity levels. On R + 28, 1 subject who reported low back pain did not complete a visual analogue scale. BDC, day of pre-bed-rest baseline data collection.

subjects ( $P = 0.016$ ) with no effects seen for the erector spinae or quadratus lumborum ( $P \geq 0.12$ ).

*Changes in muscle signal intensity with loading.* After bed rest, but not beforehand, significant increases in signal intensity in the of the erector spinae (R + 1:  $P = 0.018$  and R + 5:

$P = 0.031$ ) and multifidus (R + 1:  $P = 0.018$ ; Table 5) were seen at the 30% body-weight force level. ANOVA showed, however, that these effects over time were not statistically significant ( $P \geq 0.26$ ). Similarly, in the psoas and quadratus lumborum muscles no significant effects were seen for the

Table 2. Baseline disc and spinal morphology at each vertebral level

Parameter	Vertebral Level					
	T12L1	L1L2	L2L3	L3L4	L4L5	L5S1
Disc volume, cm <sup>3</sup>	8.8 (2.4)	10.8 (2.2)	12.3 (2.2)	12.1 (2.2)	10.1 (2.2)	6.4 (2.2)
Intervertebral length, mm	31.3 (1.8)	33.7 (1.8)	34.4 (1.7)	34.7 (1.8)	33.2 (1.7)	26.8 (1.9)
Anterior disc height, mm	7.2 (1.4)	8.6 (1.4)	9.2 (1.4)	9.9 (1.4)	10.1 (1.4)	10.4 (1.6)
Posterior disc height, mm	4.4 (0.8)	5.3 (0.8)	6.2 (0.8)	6.1 (0.8)	5.7 (0.8)	4.3 (0.8)

Values are mean(SD) from 1st campaign baseline data collection (BDC).



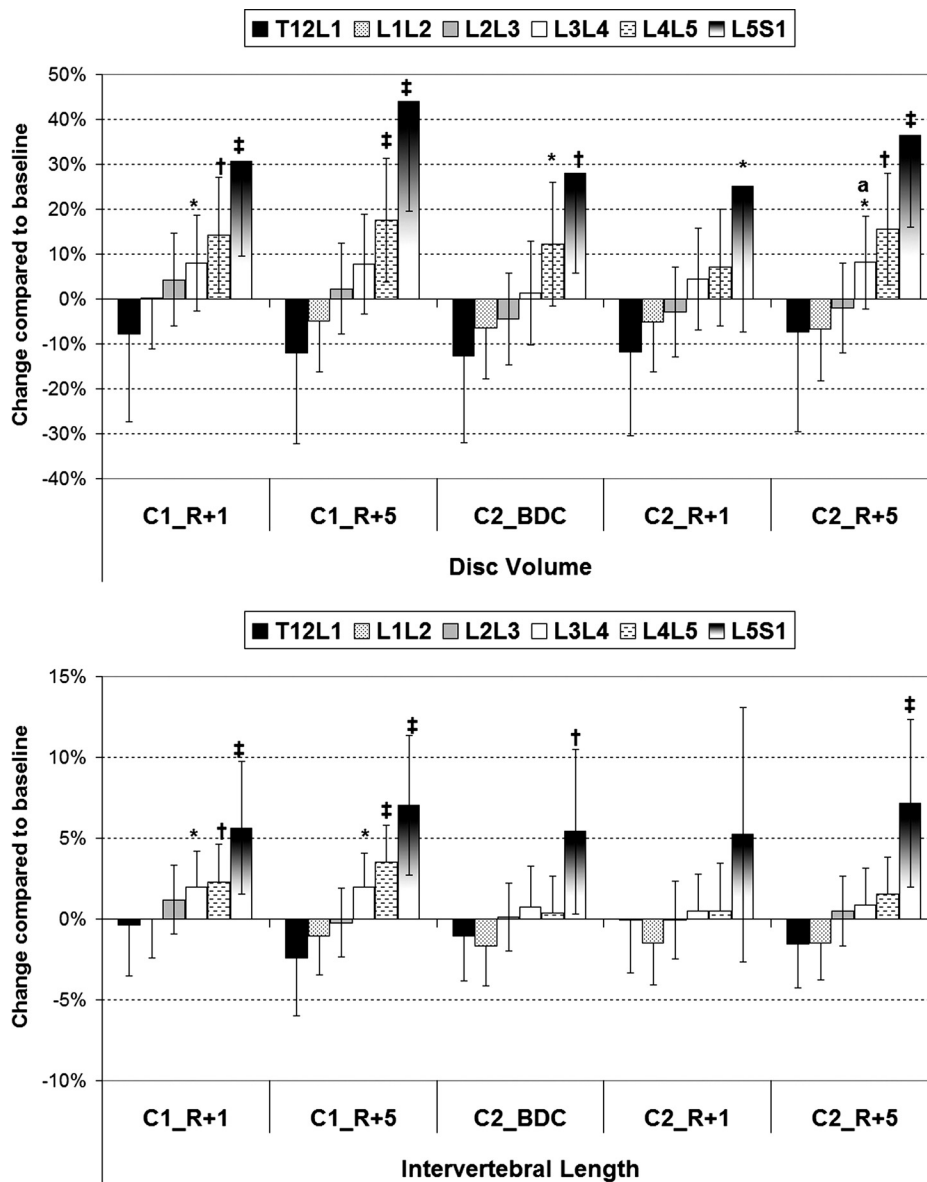


Fig. 4. Changes in disc volume (top) and intervertebral length (bottom). Values are mean(SD) percentage change compared with 1st campaign baseline (BDC). R + 1 and R + 5 represent measurements performed 1 and 5 days after the end of bed rest. C1, 1st campaign; C2, 2nd campaign. The 2nd campaign BDC occurred 147 days after 1st campaign R + 5, and all 7 subjects completed all scanning sessions. \* $P < 0.05$ , † $P < 0.01$ , ‡ $P < 0.001$ , significance of difference to 1st campaign baseline value. <sup>a</sup> $P < 0.05$ , significance of difference on 2nd campaign R + 1 and R + 5 to 2nd campaign BDC. A significant study-date  $\times$  vertebral-level interaction was seen for disc volume and intervertebral length (see RESULTS for further details). Note that increased lower lumbar spine disc volume and intervertebral length seen at R + 5 in the 1st campaign persist 147 days later at 2nd campaign BDC (equivalent to R + 153).

change in signal intensity due to loading over the course of the study ( $P \geq 0.14$ ; Table 5). Analysis of signal intensity changes with loading at each vertebral level did not show any significant changes on ANOVA over the course of the study (data not shown). Subjects reporting low back pain between R + 0 and R + 28 showed a higher level of multifidus signal intensity change (both loading levels pooled) on R + 1 than those that did not report low back pain ( $P = 0.038$ ). The increases in multifidus muscle activation with loading compared with before bed rest did not quite reach significance, however (subjects reporting low back pain: +6.0%;  $P = 0.062$ ; subject not reporting low back pain: +0.1%;  $P = 0.70$ ). In terms of low back pain reports after bed rest, no differences were observed for the response of signal intensity in the other muscles ( $P \geq 0.13$ ).

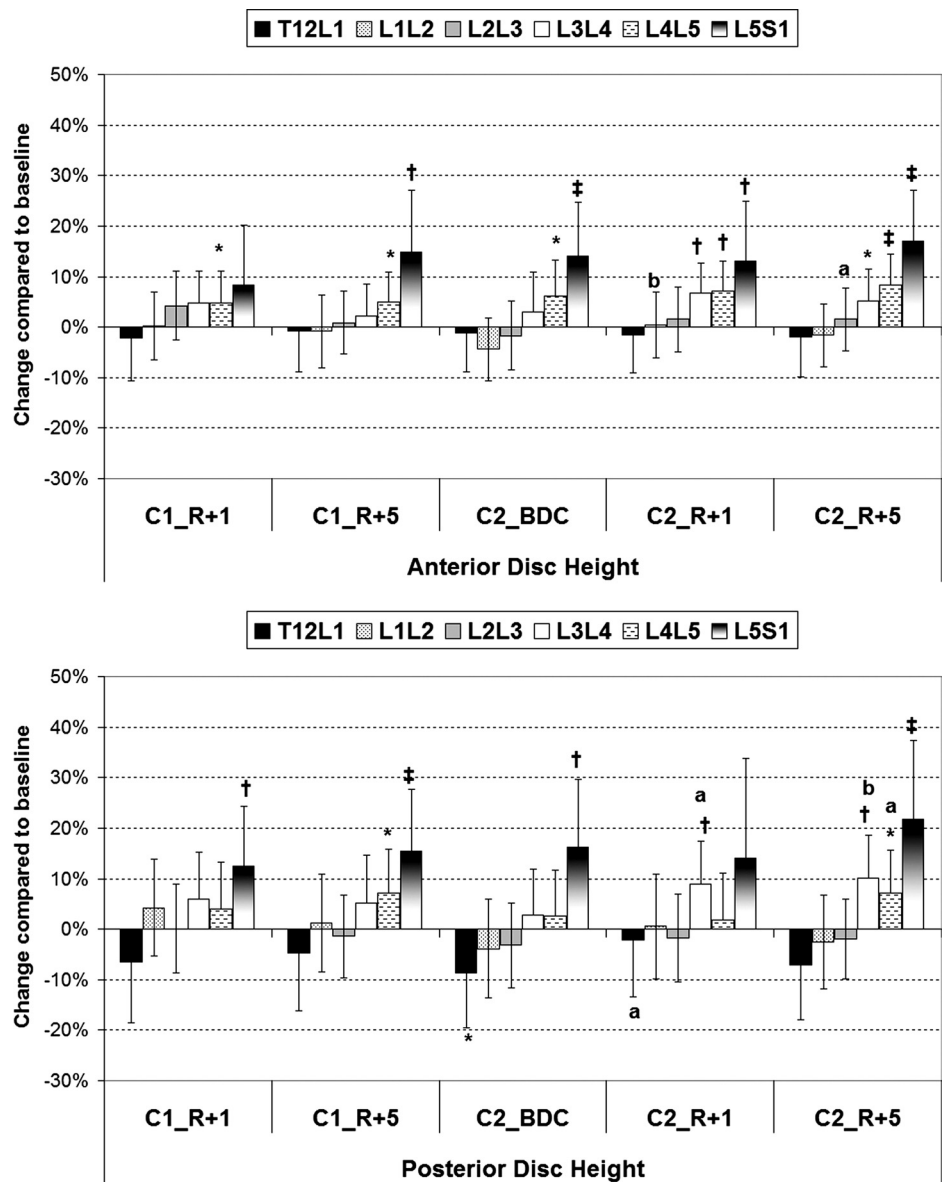
## DISCUSSION

The main finding of the current study was that the lumbar intervertebral discs did not return to their prebed-rest state 153

days after 21-day bed rest. This effect was apparent for anterior and posterior disc height, disc volume, and intervertebral length and was particularly evident at the lower lumbar spine. Muscle CSA recovery did occur in this time frame, and for some lumbar muscles, CSA was seen to be significantly greater at this recovery time point than prebed-rest values. The extent of muscle CSA changes and pattern of changes in disc and spine morphology due to bed rest are largely consistent with prior work (2–4, 6, 14, 17, 21, 28, 29, 31, 44).

That disc remodeling could be protracted after bed rest is not unreasonable, given that for other tissues, such as bone (43), the duration required to rebuild losses during bed rest is a number orders longer than the time required to lose it. While other studies (6, 19, 21, 28) have evaluated the intervertebral discs in recovery, the current work is the first to date to examine the intervertebral discs this late into recovery. Overall, the available data suggest the recovery process of the intervertebral discs after bed rest is indeed protracted. While animal data from disc immobilization (50), hindlimb unloading (20,

Fig. 5. Changes in anterior (*top*) and posterior (*bottom*) disc height. Values are mean(SD) percentage change compared with 1st campaign baseline (BDC). R+1 and R+5 represent measurements performed 1 and 5 days after the end of bed rest. The 2nd campaign BDC occurred 147 days after 1st campaign R + 5, and all 7 subjects completed all scanning sessions.  $P < 0.05$ ,  $^{\dagger}P < 0.01$ ,  $^{\ddagger}P < 0.001$ , significance of difference to 1st campaign baseline value.  $^aP < 0.05$ ,  $^bP < 0.01$ , significance of difference on 2nd campaign R + 1 and R + 5 to 2nd campaign BDC. Study-date  $\times$  vertebral-level interaction was significant for these two variables (see RESULTS for further details). Note that increased anterior and posterior disc height seen at R + 5 in the 1st campaign persist 147 days later at 2nd campaign BDC (equivalent to R + 153).



22), or microgravity (33, 45) suggest proteoglycan content reduces in the intervertebral discs with a reduction of disc anabolism but increase in catabolism (32, 50), similar data are not available in humans. One animal study (50) showed proteoglycan content was not completely recovered 3 wk after 3 wk of disc immobilization. Due to the mechanical differences between animals and humans in the role of the lumbar spine in locomotion, caution needs to be exercised in relating the finding of animal studies to those in humans. Where possible, examination of such metabolic/biochemical disc parameters in humans could provide greater insight into the mechanisms at play in the protracted recovery process. Data available from human tissue on aggrecan (the most common proteoglycan in the intervertebral disc; Ref. 24) and collagen turnover in the intervertebral disc suggest that the “half-life” (time until 50% is turned over) for aggrecan to be  $\sim 5$  yr (46) and  $\sim 95$  yr for collagen (47). These data suggest the remodeling of the disc is slow process. Overall, the available data suggest that the recovery of the intervertebral disc after bed rest is indeed

protracted and future work should evaluate the long-term recovery, along with consideration of other parameters, such as proteoglycan content as well as evaluating the nucleus pulposus and annulus fibrosus separately. Another relevant question for future work would be what duration of bed rest is necessary before such a protracted, and potentially incomplete, recovery process is to be expected.

Could these findings from bed rest on the intervertebral discs be clinically relevant? Muscle atrophy and loss of bone during bed rest are considered to be negative effects of bed rest that are to be prevented, but is the same true of the increases in disc height/volume seen in the current study? While loss of disc height is commonly associated with age and disc degeneration (39), readers should be cautioned against naively assuming that the increases of disc height seen in the current study represent a “beneficial” effect: the physiological processes and changes in disc tissue structure associated with degeneration are unlikely to be the same as those occurring during disc unloading. Data from biomechanical modeling studies help, however, to

Table 3. Lumbar muscle cross-sectional area at baseline and changes throughout the study

Muscle and Vertebral Level	Study Date					
	1st Campaign			2nd Campaign		
	BDC, cm <sup>2</sup>	R + 1, %	R + 5, %	BDC, %	R + 1, %	R + 5, %
Erector Spinae						
L1	17.1 (1.9)	-9.7 (4.5)%‡	-7.7 (5.0)%‡	0.9 (5.3)%	-5.3 (5.2)% <sup>†b</sup>	-5.4 (5.0)% <sup>†b</sup>
L2	19.0 (1.9)	-8.3 (4.1)%‡	-6.1 (4.1)%‡	1.7 (4.4)%	-6.3 (4.4)% <sup>‡c</sup>	-3.6 (4.1)% <sup>*b</sup>
L3	18.4 (1.9)	-6.1 (4.0)%‡	-3.5 (4.0)%*	3.1 (4.7)%	-4.1 (3.7)% <sup>†c</sup>	-1.6 (4.0)% <sup>b</sup>
L4	15.6 (2.0)	-4.6 (5.8)%*	-1.5 (5.6)%	2.3 (6.6)%	-5.0 (6.0)% <sup>*b</sup>	-1.8 (5.4)%
L5	11.2 (2.1)	-3.4 (7.8)%	-3.8 (8.5)%	2.3 (10.1)%	2.0 (13.6)%	1.9 (7.8)%
Multifidus						
L1	2.4 (0.5)	-8.4 (7.8)% <sup>†</sup>	-7.8 (6.5)% <sup>†</sup>	1.8 (6.5)%	-3.7 (6.2)% <sup>b</sup>	-1.6 (6.2)% <sup>a</sup>
L2	2.9 (0.5)	-5.6 (8.1)%	-4.9 (6.3)%*	3.5 (6.6)%	-3.8 (6.4)% <sup>b</sup>	-1.6 (6.1)% <sup>a</sup>
L3	4.0 (0.5)	-2.0 (5.2)%	0.4 (4.8)%	4.9 (5.4)%*	-2.5 (4.4)% <sup>c</sup>	0.4 (5.5)% <sup>a</sup>
L4	5.2 (0.5)	-5.5 (4.9)% <sup>†</sup>	1.1 (4.2)%	6.1 (6.0)% <sup>†</sup>	-3.6 (5.4)% <sup>c</sup>	1.5 (4.8)% <sup>a</sup>
L5	6.5 (0.6)	-6.5 (7.0)%*	1.8 (6.0)%	8.1 (6.9)% <sup>†</sup>	-8.4 (6.6)% <sup>†c</sup>	1.3 (5.6)%
Psoas						
L1	1.5 (1.5)	-2.1 (24.3)%	-7.5 (23.4)%	13.2 (24.0)%	15.8 (23.2)%	11.2 (23.3)%
L2	6.3 (1.5)	-0.3 (5.8)%	-1.9 (5.4)%	0.5 (5.2)%	0.0 (5.1)%	1.2 (5.5)%
L3	11.2 (1.5)	1.5 (3.7)%	2.3 (3.4)%	1.8 (4.2)%	-0.4 (3.8)%	1.3 (3.7)%
L4	15.4 (1.6)	3.6 (5.6)%	6.4 (4.7)%‡	3.2 (7.1)%	-1.1 (6.4)%	2.9 (5.4)%
L5	15.5 (1.7)	6.6 (8.6)%*	13.2 (8.0)%‡	10.1 (8.3)% <sup>†</sup>	-1.5 (10.5)% <sup>b</sup>	9.7 (7.3)%‡
Quadratus Lumborum						
L1	1.7 (1.4)	-7.0 (15.6)%	-6.7 (14.7)%	-2.2 (15.4)%	-5.7 (14.9)%	-3.1 (16.8)%
L2	4.2 (1.4)	-5.2 (6.3)%*	-3.4 (6.1)%	0.5 (6.7)%	-6.0 (5.7)% <sup>†c</sup>	-2.8 (5.8)%
L3	5.6 (1.4)	-4.8 (4.6)% <sup>†</sup>	-1.5 (4.6)%	-0.1 (5.2)%	-5.9 (4.9)% <sup>†b</sup>	-3.5 (4.4)% <sup>*b</sup>
L4	7.0 (1.4)	1.1 (4.5)%	4.5 (4.1)% <sup>†</sup>	5.5 (5.6)%*	-0.5 (5.2)% <sup>a</sup>	1.3 (4.1)% <sup>a</sup>
L5	—	—	—	—	—	—

Values are mean(SD): at baseline (BDC) in the 1st campaign in cm<sup>2</sup> and subsequently in percentage change compared with 1st campaign baseline. R + 1 and R + 5 are measurements performed 1 and 5 days after the end of bed rest. The 2<sup>nd</sup> campaign BDC occurred 147 days after 1<sup>st</sup> campaign R + 5, and all 7 subjects completed all scanning sessions. \* $P < 0.05$ ,  $^†P < 0.01$ ,  $^‡P < 0.001$ , significance of difference to 1st campaign baseline value. <sup>a</sup> $P < 0.05$ , <sup>b</sup> $P < 0.01$ , <sup>c</sup> $P < 0.001$ , significance of difference on 2nd campaign R + 1 and R + 5 to 2nd campaign BDC.

understand the findings of the current study. These studies have shown reductions of disc stiffness (36), increased zygapophyseal joint compressive load (51), increased intradiscal pressure (51), increased longitudinal stresses in the posterior portion of the disc (30), increased disc bulging (30), and increased axial disc displacement (30) when loading of discs of greater height is performed. These biomechanical changes imply decreased intersegmental stiffness, which would need to be controlled by

Table 4. Partial correlation coefficients between changes in muscle size and disc and spine morphology parameters

Parameter	Disc Volume	Anterior Disc Height	Posterior Disc Height	Intervertebral Length
Erector Spinae	0.57	0.71	0.82	0.44
Multifidus	0.76	0.88	0.69	0.68
Psoas	0.69	0.70	0.21	0.66
Quad. Lumborum	0.94	0.84	0.91	0.95

Values are Pearson's partial correlation coefficient (controlling for study-date) based on percentage change in each parameter on R + 1 compared with before bed rest. Data averaged across the entire lumbar spine have been used. Results are similar when correlations are performed on data from each individual vertebral level. Data are from 1st campaign only. When 2nd campaign data are included in analysis, correlations reduce for the erector spinae muscle but not for the other muscles (data not shown). The aim of this correlation analysis was to examine the relationship between the changes in the variables to aid in the interpretation of the data, rather than in assessing significance of these relationships per se. Nonetheless, given an  $n = 7$ , the  $P$  value without Bonferroni correction reaches  $<0.05$  when the correlation is  $>0.75$ . Positive correlations between changes in muscle cross-sectional area and changes in disc and spine morphology imply that decreases of muscle cross-sectional area are unlikely to be causally associated with increases of disc volume/height and spinal length.

the (deconditioned) muscle system (37, 38). There is also some clinical data available to help understand the potential implications of the current findings: 1) greater disc height is a risk factor for recurrence of disc herniation (25); 2) the available data on the time of onset of acute low back pain suggest increased incidence before midday (48) when the intervertebral discs are still reducing in size after overnight increases as part of normal diurnal variation (12, 13); 3) in astronauts, increased incidence of lumbar intervertebral disc prolapse after spaceflight has been observed (23), although there is insufficient data on morphological changes in the intervertebral discs after spaceflight to relate this to prolapse incidence, and 4) prior work (3, 21), but not the current study, has shown an association between greater increases in disc volume or height during bed rest and the incidence of low back pain after bed rest. It is worth keeping in mind that the persistence of the changes in the discs may also be due to changes in other structures or altered loading patterns. In any case, the partial correlations analyses between disc changes and muscle changes suggest that the alterations of muscle CSA are not related to the persistence of the changes in the discs. Overall, the persistent changes in the intervertebral disc observed after prolonged bed rest may indeed have negative clinical consequences, although this relationship needs to be investigated further.

In terms of the musculature, CSA recovered within the time frame considered. This recovery process appears to take some time, however, with the current and other available (4, 6, 34, 43) data suggesting that muscle CSA recovery occurs within 3 mo after bed rest. It should be noted, however, that data are available suggesting the recovery of lumbar muscle motor



Table 5. Muscle signal intensity with isometric spinal extension exercise

Muscle and Loading Condition	Study Date					
	10% Body Weight			30% Body Weight		
	BDC	R + 1	R + 5	BDC	R + 1	R + 5
Erector Spinae						
Before	84.0 (7.3)	80.7 (7.7)	83.2 (7.5)	82.7 (8.0)	81.1 (8.2)	81.2 (7.7)
After	0.6 (3.2)%	2.1 (7.6)%	-0.3 (4.6)%	0.5 (9.1)%	7.9 (7.6)%*	5.1 (5.5)%*
Multifidus						
Before	89.4 (7.5)	85.7 (7.9)	88.3 (7.7)	88.1 (8.8)	89.0 (9.0)	87.7 (7.8)
After	0.6 (4.7)%	1.2 (7.9)%	-0.7 (5.8)%	0.3 (17.0)%	9.2 (8.9)%*	1.5 (6.4)%
Psoas						
Before	46.9 (4.4)	45.2 (4.5)	46.1 (4.4)	47.6 (4.8)	46.9 (4.5)	45.9 (4.4)
After	1.8 (5.2)%	1.5 (8.3)%	0.8 (5.5)%	-0.1 (9.6)%	5.2 (6.8)%	1.0 (5.3)%
Quadratus lumborum						
Before	55.9 (5.0)	55.8 (5.0)	57.3 (4.1)	57.9 (6.2)	54.1 (7.1)	55.7 (6.2)
After	4.7 (4.7)%*	1.2 (7.4)%	-0.9 (5.5)%	2.0 (9.8)%	7.9 (11.3)%	5.7 (7.1)%

Values are mean(SD). Values from "before" exercise are signal intensity in absolute values (no units) and "after" loading, percentage changes in signal intensity are given. No significant differences in "before" signal intensity between study dates. Signal intensity was averaged between vertebral levels, weighted by cross-sectional area, before further analysis. Note: due to organizational and time constraints, 10% body weight loading was performed in 1st campaign and 30% body weight performed in 2nd campaign. \* $P < 0.05$ , significant increase in signal intensity occurred after exercise. While signal intensity increase differed after bed rest for the 30% body weight loading level in the erector spinae and multifidus, ANOVA suggested that these effects were not significant ( $P \geq 0.14$ ).

control after bed rest takes much longer (5, 7, 8). Caution needs to be applied when interpreting the increases in muscle CSA from R + 1 to R + 5 as "recovery." This finding is likely not due to solely to muscle fiber recovery, and a component may be associated with muscle swelling associated with delayed onset muscle soreness and injury of atrophied muscle after bed rest (41). Overall, given the role of the musculature in stabilizing the lumbar spine (37), the data suggest there is likely a time window of higher injury risk after bed rest as a consequence of muscular deconditioning and changes in disc and spine morphology.

Interestingly, muscle CSA was indeed larger ~5 mo after the first campaign than before bed rest in some muscles. Of the few publications that also present data from late in the recovery phase similar effects have been seen for some muscles (6, 19, 34, 43). What could be behind these effects? Subjects typically receive either no rehabilitation or only a general rehabilitation program but not specific resistive exercise protocols, which are best known to increase muscle size. Therefore, is participation in bed rest, in and of itself, in the long-run "beneficial" for the musculature? Prior work (9) has shown an increase in intramuscular connective tissue after bed rest associated with muscle fiber atrophy. It could be that muscles become "larger" in the long-run after bed rest as muscle fiber recovers after bed rest, but connective tissue may not reduce to prebed rest levels, resulting in increased CSA. An alternative interpretation is that the increased intervertebral distance after bed rest requires increased muscular torque development for trunk stabilization, which could potentially constitute a stimulus for muscle hypertrophy. In line with this interpretation the partial correlation coefficients in Table 4 between muscle and disc changes were always positive. However, future work would need to address this question specifically by evaluating the separate components of muscle (connective tissue, muscle fiber, fluid content, and fat content). It is worth noting that psoas muscle CSA actually increased after bed rest, something not observed after spaceflight (27). This effect on psoas in bed rest has been observed in other studies (3, 17, 44) and stresses that bed rest

is not necessarily a model of inactivity, or spaceflight, for all muscles or body systems.

In the current study, a secondary goal was also to conduct a pilot study examining lumbar muscle activation as measured by signal intensity changes before and after isometric spinal extension. While the effects were not significant on ANOVA, greater increases in signal intensity were seen in the erector spinae and multifidus after bed rest compared with before bed rest. These data could indicate greater activation of this muscle group to generate the forces necessary during the standardized loading task in the face of muscle atrophy. While this pilot work on signal intensity changes with exercise shows some promise as an outcome measure for examining lumbar muscle activation after bed rest, further refinement of the measurement methodology would be necessary to define and improve repeatability for use in the small subject pools of bed rest studies. Also, measurement of  $T_2$  relaxation time may provide more specific data on muscle water content that is less subject to changes in MR field homogeneity.

Interestingly, however, there was some indication that subjects who reported low back pain after bed rest showed greater activation of the multifidus muscle during the standardized loading task. The multifidus muscle is, from a biomechanical point of view (10, 49), particularly important for stabilizing the lumbar spine. This is underscored by the finding of the current and prior (3) work that the extent of multifidus muscle atrophy due to bed rest was associated with the incidence of low back pain after bed rest.

It is also worth considering some of the limitations of the current study. Due to restricted access to MR facility, scanning could not be done in the bed rest phase and subjects were first scanned again 1 day after bed rest. Since the current study focused on the recovery phase, this is not a major limitation, but the reader should be aware that the values on R + 1 may not be the same as those seen at the end of bed rest before the subjects reambulated. Also, in the current work, it is more difficult to implicate the MR changes on R + 1 as "causes" of low back pain between R + 0 and R + 28 as they could

potentially be “effects” of low back pain on R + 0 and R + 1 (there were three reports of low back pain on R + 0 and two on R + 1). Furthermore, as is common in bed rest, the number of subjects was restricted due to logistical and financial restraints. It could be that some statistically nonsignificant findings represent false-negatives. Also, due to the small sample size, it is possible that some significant effects may have been detected simply because this small collective behaved differently to what may have been seen in a larger collective. An example of this is that in the current study significant differences were seen between the lumbar intervertebral discs in terms of volume changes, which is something not seen in other studies (3, 6), although this finding on the intervertebral discs could be related to the, compared with prior work, younger collective in the current study.

In conclusion, the current study found that lumbar multifidus, psoas, and quadratus lumborum muscle CSA was significantly larger 153 days after 21-day bed rest than at baseline testing. Further work will need to examine whether this represents muscle fiber hypertrophy or changes in other intramuscular structures. A second bout of 21-day bed rest in the same subjects resulted in a similar extent of muscle CSA and spinal morphology changes as after the first bout. The report of low back pain after bed rest was associated with greater reductions of multifidus and psoas CSA and greater activation of the multifidus muscle, as indicated by signal intensity changes, in a standardized isometric spinal extension loading task. The main finding of the current study was, however, that the lumbar intervertebral discs did not return to their prebed rest state 153 days after 21-day bed rest. While there are some indications from other studies that these effects may have negative clinical consequences, further work needs to evaluate this relationship as well as the long-term time course of intervertebral disc recovery.

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#### DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author(s).

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